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Terms	Documents
L2 and (424/450).ccls.	28

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DATE: Thursday, March 24, 2005 [Printable Copy](#) [Create Case](#)

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<u>L3</u>	L2 and 424/450.ccls.	28	<u>L3</u>
<u>L2</u>	liposome same cholate	80	<u>L2</u>
<u>L1</u>	liposome same cholate same antibody	3	<u>L1</u>

END OF SEARCH HISTORY

Hit List

Clear

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Fwd Refs

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Search Results - Record(s) 1 through 28 of 28 returned.

☐ 1. Document ID: US 6726924 B2

Using default format because multiple data bases are involved.

L3: Entry 1 of 28

File: USPT

Apr 27, 2004

US-PAT-NO: 6726924

DOCUMENT-IDENTIFIER: US 6726924 B2

TITLE: Oral liposomal delivery system

DATE-ISSUED: April 27, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Keller; Brian C.	Antioch	CA		

US-CL-CURRENT: [424/450](#); [424/451](#), [424/452](#), [424/453](#), [424/455](#), [424/456](#)

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw. D.
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☐ 2. Document ID: US 6723338 B1

L3: Entry 2 of 28

File: USPT

Apr 20, 2004

US-PAT-NO: 6723338

DOCUMENT-IDENTIFIER: US 6723338 B1

**** See image for [Certificate of Correction](#) ****

TITLE: Compositions and methods for treating lymphoma

DATE-ISSUED: April 20, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Sarris; Andreas H.	Houston	TX		
Cabanillas; Fernando	Houston	TX		
Logan; Patricia M.	Vancouver			CA
Burge; Clive T. R.	Brentwood Bay			CA
Goldie; James H.	Vancouver			CA
Webb; Murray S.	Delta			CA

US-CL-CURRENT: [424/450](#)

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWC	Draw. D.
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☐ 3. Document ID: US 6713533 B1

L3: Entry 3 of 28

File: USPT

Mar 30, 2004

US-PAT-NO: 6713533

DOCUMENT-IDENTIFIER: US 6713533 B1

TITLE: Nanocapsules and method of production thereof

DATE-ISSUED: March 30, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Panzner; Steffen	Halle			DE

US-CL-CURRENT: 523/202; 424/450, 424/451, 524/205, 524/210

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWC	Draw. D.
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☐ 4. Document ID: US 6706861 B2

L3: Entry 4 of 28

File: USPT

Mar 16, 2004

US-PAT-NO: 6706861

DOCUMENT-IDENTIFIER: US 6706861 B2

TITLE: Reconstitution of purified membrane proteins into preformed liposomes

DATE-ISSUED: March 16, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Singh; Pratap	Wilmington	DE		
Wang; Jianfang	Wilmington	DE		
Tejidor; Liliana Maria	Raleigh	NC		

US-CL-CURRENT: 530/381; 424/450, 435/13, 436/69, 436/829, 530/350, 530/380

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWC	Draw. D.
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☐ 5. Document ID: US 6451543 B1

L3: Entry 5 of 28

File: USPT

Sep 17, 2002

US-PAT-NO: 6451543

DOCUMENT-IDENTIFIER: US 6451543 B1

TITLE: Lipid matrix-assisted chemical ligation and synthesis of membrane polypeptides

DATE-ISSUED: September 17, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Kochendoerfer; Gerd. G.	Oakland	CA		
Hunter; Christie L.	San Francisco	CA		
Kent; Stephen B. H.	San Francisco	CA		
Botti; Paolo	San Francisco	CA		

US-CL-CURRENT: 435/7.1; 424/450, 435/870, 436/501, 436/544, 436/87, 436/88, 436/89, 436/90, 530/334, 530/359, 530/400, 530/402, 530/404, 530/408

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw D
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☐ 6. Document ID: US 6443898 B1

L3: Entry 6 of 28

File: USPT

Sep 3, 2002

US-PAT-NO: 6443898

DOCUMENT-IDENTIFIER: US 6443898 B1

TITLE: Therapeutic delivery systems

DATE-ISSUED: September 3, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Unger; Evan C.	Tucson	AZ		
Fritz; Thomas A.	Tucson	AZ		
Matsunaga; Terry	Tucson	AZ		
Ramaswami; VaradaRajan	Tucson	AZ		
Yellowhair; David	Tucson	AZ		
Wu; Guanli	Tucson	AZ		

US-CL-CURRENT: 600/458; 424/450, 424/9.51

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw D
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☐ 7. Document ID: US 6372720 B1

L3: Entry 7 of 28

File: USPT

Apr 16, 2002

US-PAT-NO: 6372720

DOCUMENT-IDENTIFIER: US 6372720 B1

TITLE: Liposome fusion and delivery vehicle

DATE-ISSUED: April 16, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Longmuir; Kenneth J.	Irvine	CA	92612	
Waring; Alan J.	Irvine	CA	92614	
Haynes; Sherry M.	Irvine	CA	92612	

US-CL-CURRENT: 514/44; 424/450, 435/320.1, 435/455, 435/458, 514/2

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWAC	Draw. De
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☐ 8. Document ID: US 6287590 B1

L3: Entry 8 of 28

File: USPT

Sep 11, 2001

US-PAT-NO: 6287590

DOCUMENT-IDENTIFIER: US 6287590 B1

**** See image for Certificate of Correction ****

TITLE: Peptide/lipid complex formation by co-lyophilization

DATE-ISSUED: September 11, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Dasseux; Jean-Louis	Mannheim			DE

US-CL-CURRENT: 424/450; 514/12, 514/13, 514/2

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWAC	Draw. De
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☐ 9. Document ID: US 6248353 B1

L3: Entry 9 of 28

File: USPT

Jun 19, 2001

US-PAT-NO: 6248353

DOCUMENT-IDENTIFIER: US 6248353 B1

TITLE: Reconstitution of purified membrane proteins into preformed liposomes

DATE-ISSUED: June 19, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Singh; Pratap	Wilmington	DE		

US-CL-CURRENT: 424/450; 424/94.3, 436/829, 530/350, 530/381

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWAC	Draw. De
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☐ 10. Document ID: US 6019998 A

L3: Entry 10 of 28

File: USPT

Feb 1, 2000

US-PAT-NO: 6019998

DOCUMENT-IDENTIFIER: US 6019998 A

**** See image for Certificate of Correction ****

TITLE: Membrane structure

DATE-ISSUED: February 1, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Nomoto; Tsuyoshi	Atsugi			JP
Tomida; Yasuko	Atsugi			JP
Ohyama; Junji	Yamato			JP
Maruyama; Tomoko	Atsugi			JP

US-CL-CURRENT: 424/450; 264/4.32, 264/4.33, 264/4.7, 427/213.33, 427/213.34,
427/214, 428/432, 428/433, 428/470

Full	Title	Citation	Front	Review	Classification	Date	Reference				Claims	KWIC	Draw D
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☐ 11. Document ID: US 5952312 A

L3: Entry 11 of 28

File: USPT

Sep 14, 1999

US-PAT-NO: 5952312

DOCUMENT-IDENTIFIER: US 5952312 A

TITLE: NADH and NADPH therapeutic agents for nasal, sublingual, rectal and dermal administration

DATE-ISSUED: September 14, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Birkmayer; Joerg G. D.	Vienna			AT

US-CL-CURRENT: 514/47; 424/401, 424/450

Full	Title	Citation	Front	Review	Classification	Date	Reference				Claims	KWIC	Draw D
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☐ 12. Document ID: US 5948756 A

L3: Entry 12 of 28

File: USPT

Sep 7, 1999

US-PAT-NO: 5948756

DOCUMENT-IDENTIFIER: US 5948756 A

TITLE: Therapeutic lipoprotein compositions

DATE-ISSUED: September 7, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Barenholz; Yechezkel	Jerusalem			IL
Shmeeda; Hilary	Givat Zev			IL
Chajek; Tova	Jerusalem			IL

US-CL-CURRENT: 514/12; 424/417, 424/450, 424/460, 514/2, 514/7, 514/789, 530/352,
530/359

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	RMK	Draw Ds
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☐ 13. Document ID: US 5914311 A

L3: Entry 13 of 28

File: USPT

Jun 22, 1999

US-PAT-NO: 5914311

DOCUMENT-IDENTIFIER: US 5914311 A

TITLE: Method for reducing serum lipoprotein (A) concentration

DATE-ISSUED: June 22, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Barenholz; Yechezkel	Jerusalem			IL
Shmeeda; Hilary	Givat Zev			IL
Chajek; Tova	Jerusalem			IL

US-CL-CURRENT: 514/1; 424/450, 514/12

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	RMK	Draw Ds
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☐ 14. Document ID: US 5910306 A

L3: Entry 14 of 28

File: USPT

Jun 8, 1999

US-PAT-NO: 5910306

DOCUMENT-IDENTIFIER: US 5910306 A

TITLE: Transdermal delivery system for antigen

DATE-ISSUED: June 8, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Alving; Carl R.	Bethesda	MD		
Glenn; Gregory M.	Bethesda	MD		

US-CL-CURRENT: 424/184.1; 424/204.1, 424/234.1, 424/265.1, 424/269.1, 424/274.1,
424/277.1, 424/279.1, 424/282.1, 424/283.1, 424/449, 424/450, 424/810, 424/812

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KIMC	Draw. De
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☐ 15. Document ID: US 5908777 A

L3: Entry 15 of 28

File: USPT

Jun 1, 1999

US-PAT-NO: 5908777

DOCUMENT-IDENTIFIER: US 5908777 A

TITLE: Lipidic vector for nucleic acid delivery

DATE-ISSUED: June 1, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lee; Robert J.	Pittsburgh	PA		
Huang; Leaf	Wexford	PA		

US-CL-CURRENT: 435/320.1; 264/4.1, 424/450, 424/93.21, 435/325, 435/458, 435/69.1,
514/44

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KIMC	Draw. De
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☐ 16. Document ID: US 5863556 A

L3: Entry 16 of 28

File: USPT

Jan 26, 1999

US-PAT-NO: 5863556

DOCUMENT-IDENTIFIER: US 5863556 A

**** See image for Certificate of Correction ****

TITLE: Preparations for the external application of antiseptic agents and/or agents promoting the healing of wounds

DATE-ISSUED: January 26, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Ruckert; Dieter	Tubingen			DE
Gumbel; Herman	Rodermark-Waldacker			DE
Fleischer; Wolfgang	Ingelheim			DE
Reimer; Karen	Limburg			DE
Winkler; Horst	Limburg			DE

US-CL-CURRENT: 424/450; 424/405, 424/417, 424/78.04, 424/78.07, 424/78.24

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWNC	Draw D
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☐ 17. Document ID: US 5853753 A

L3: Entry 17 of 28

File: USPT

Dec 29, 1998

US-PAT-NO: 5853753

DOCUMENT-IDENTIFIER: US 5853753 A

**** See image for Certificate of Correction ****

TITLE: Liposomes, method of preparing the same and use thereof in the preparation of drugs

DATE-ISSUED: December 29, 1998

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Maierhofer; Gunther	Munich			DE
Hofer; Paul	Dietersheim			DE
Rottmann; Oswald	Freising			DE

US-CL-CURRENT: 424/450

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWNC	Draw D
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☐ 18. Document ID: US 5591448 A

L3: Entry 18 of 28

File: USPT

Jan 7, 1997

US-PAT-NO: 5591448

DOCUMENT-IDENTIFIER: US 5591448 A

TITLE: Anti-viral therapeutic composition

DATE-ISSUED: January 7, 1997

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Tepic; Slobodan	CH-7270 Davos			CH

US-CL-CURRENT: 424/450; 428/402.2, 436/829

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWNC	Draw D
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☐ 19. Document ID: US 5543399 A

L3: Entry 19 of 28

File: USPT

Aug 6, 1996

US-PAT-NO: 5543399

DOCUMENT-IDENTIFIER: US 5543399 A

**** See image for Certificate of Correction ****

TITLE: Cystic fibrosis transmembrane conductance regulator (CFTR) protein

DATE-ISSUED: August 6, 1996

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Riordan; John R.	Toronto			CA
Bear; Christine E.	North York			CA
Ramjeesingh; Mohabir	Mississauga			CA
Li; Canhui	Toronto			CA

US-CL-CURRENT: 514/21; 424/450, 514/12, 514/8, 514/851, 530/350, 530/415, 530/417

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw. De
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☐ 20. Document ID: US 5542935 A

L3: Entry 20 of 28

File: USPT

Aug 6, 1996

US-PAT-NO: 5542935

DOCUMENT-IDENTIFIER: US 5542935 A

**** See image for Certificate of Correction ****

TITLE: Therapeutic delivery systems related applications

DATE-ISSUED: August 6, 1996

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Unger; Evan C.	Tucson	AZ		
Fritz; Thomas A.	Tucson	AZ		
Matsunaga; Terry	Tucson	AZ		
Ramaswami; VaradaRajan	Tucson	AZ		
Yellowhair; David	Tucson	AZ		
Wu; Guanli	Tucson	AZ		

US-CL-CURRENT: 604/190; 424/450, 600/458

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw. De
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☐ 21. Document ID: US 5328628 A

L3: Entry 21 of 28

File: USPT

Jul 12, 1994

US-PAT-NO: 5328628

DOCUMENT-IDENTIFIER: US 5328628 A

**** See image for Certificate of Correction ****

TITLE: Detergent compositions containing liposomes and process therefor

DATE-ISSUED: July 12, 1994

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hart; Gerald L.	Surbiton			GB
Ahmed; Anjum F.	Virginia Water			GB
Charaf; Ursula K.	Racine	WI		

US-CL-CURRENT: 510/418; 264/4.3, 424/450, 428/402.2, 510/122, 510/123, 510/127,
510/158, 510/431, 510/468, 514/881

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KNOW	Draw Da
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☐ 22. Document ID: US 5152999 A

L3: Entry 22 of 28

File: USPT

Oct 6, 1992

US-PAT-NO: 5152999

DOCUMENT-IDENTIFIER: US 5152999 A

TITLE: Liposome preparation

DATE-ISSUED: October 6, 1992

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Tokunaga; Yuji	Sanda			JP
Yamamoto; Takao	Osaka			JP
Hata; Takehisa	Nagaokakyo			JP

US-CL-CURRENT: 424/450; 552/544, 562/563, 562/576

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KNOW	Draw Da
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☐ 23. Document ID: US 4921757 A

L3: Entry 23 of 28

File: USPT

May 1, 1990

US-PAT-NO: 4921757

DOCUMENT-IDENTIFIER: US 4921757 A

**** See image for Certificate of Correction ****

TITLE: System for delayed and pulsed release of biologically active substances

DATE-ISSUED: May 1, 1990

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wheatley; Margaret A.	Arlington	MA		
Langer; Robert S.	Somerville	MA		
Eisen; Herman N.	Waban	MA		

US-CL-CURRENT: 428/402.2; 264/4.3, 424/418, 424/419, 424/450, 424/485, 424/488,
424/94.3, 436/829, 514/963, 514/965

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw Ds
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☐ 24. Document ID: US 4900556 A

L3: Entry 24 of 28

File: USPT

Feb 13, 1990

US-PAT-NO: 4900556

DOCUMENT-IDENTIFIER: US 4900556 A

TITLE: System for delayed and pulsed release of biologically active substances

DATE-ISSUED: February 13, 1990

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wheatley; Margaret A.	Arlington	MA		
Langer; Robert S.	Somerville	MA		
Eisen; Herman N.	Waban	MA		

US-CL-CURRENT: 424/450; 514/963

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw Ds
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☐ 25. Document ID: US 4871488 A

L3: Entry 25 of 28

File: USPT

Oct 3, 1989

US-PAT-NO: 4871488

DOCUMENT-IDENTIFIER: US 4871488 A

TITLE: Reconstituting viral glycoproteins into large phospholipid vesicles

DATE-ISSUED: October 3, 1989

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Mannino; Raphael J.	Newtonville	NY		
Fogerite; Susan G.	Waterford	NY		

US-CL-CURRENT: 264/4.6; 264/4.3, 424/1.21, 424/450, 428/402.2, 436/829, 514/8,
514/885

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KMC	Draw D
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☐ 26. Document ID: US 4792331 A

L3: Entry 26 of 28

File: USPT

Dec 20, 1988

US-PAT-NO: 4792331

DOCUMENT-IDENTIFIER: US 4792331 A

TITLE: Device for obtaining and administering unilamellar liposomes

DATE-ISSUED: December 20, 1988

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Philippot; Jean	St Clement La Riviere			FR
Liautard; Jean-Pierre	Montpellier			FR

US-CL-CURRENT: 604/187; 264/4.6, 424/450, 428/402.2, 436/829, 604/190

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KMC	Draw D
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☐ 27. Document ID: US 4731210 A

L3: Entry 27 of 28

File: USPT

Mar 15, 1988

US-PAT-NO: 4731210

DOCUMENT-IDENTIFIER: US 4731210 A

TITLE: Process for the preparation of liposomal medicaments

DATE-ISSUED: March 15, 1988

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Weder; Hans G.	Langnau			CH
Zumbuhl; Otmar N.	Wolfenschiessen			CH
Schwendener; Reto A.	Arosa			CH
Asanger; Maximilian	Zurich			CH

US-CL-CURRENT: 264/4.3; 264/4.1, 264/4.6, 424/450, 428/402.2, 436/829

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KMC	Draw D
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☐ 28. Document ID: US 4663161 A

L3: Entry 28 of 28

File: USPT

May 5, 1987

US-PAT-NO: 4663161

DOCUMENT-IDENTIFIER: US 4663161 A

TITLE: Liposome methods and compositions

DATE-ISSUED: May 5, 1987

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Mannino; Raphael J.	Newtonville	NY	12128	
Fogerite; Susan G.	Waterford	NY	12188	

US-CL-CURRENT: 424/450; 264/4.6, 424/204.1, 424/209.1, 424/211.1, 424/812,
428/402.2, 436/829

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw. De
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Clear	Generate Collection	Print	Fwd Refs	Bkwd Refs	Generate OACS
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Terms	Documents
L2 and (424/450).ccls.	28

Display Format: [Previous Page](#)[Next Page](#)[Go to Doc#](#)

Refine Search

Search Results -

Terms	Documents
L2 and (424/450).ccls.	28

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L3

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<u>L3</u>	L2 and 424/450.ccls.	28	<u>L3</u>
<u>L2</u>	liposome same cholate	80	<u>L2</u>
<u>L1</u>	liposome same cholate same antibody	3	<u>L1</u>

END OF SEARCH HISTORY

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

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L3: Entry 9 of 28

File: USPT

Jun 19, 2001

DOCUMENT-IDENTIFIER: US 6248353 B1

TITLE: Reconstitution of purified membrane proteins into preformed liposomes

Brief Summary Text (6):

There are essentially four presently known mechanisms for incorporating, i.e., reconstituting, proteins into liposomes. See Rigaud, J- L., et al., "Liposomes as Tools for the Reconstitution of Biological Systems," p. 71-88, in Liposomes as Tools in Basic Research and Industry, ed. Philippot, J. R. and Schuber, F., CRC Press, Boca Raton, Fla. (1995). One method involves the use of an organic solvent. However, such procedures often result in the denaturation of the proteins. A second method uses mechanical means to produce large and small unilamellar vesicles from MLVs by swelling of the dry phospholipid films in excess buffer. Such mechanical means include sonication of MLVs, forcing multilamellar lipid vesicles through a French press, or cycles of freeze-thawing or dehydration-rehydration. Drawbacks with sonication include variability and inactivation of certain proteins by sonication as well as production of small liposomes. A third process involves the direct incorporation of proteins into preformed small unilamellar liposomes, also termed spontaneous incorporation. Such methods are usually catalyzed by low cholesterol or lysophospholipid concentrations. Problems with these methods include the wide size distribution of the proteoliposomes, heterogeneous distribution of the protein among the liposomes and presence of the non-phospholipid impurities, required for an effective protein incorporation, that would affect performance of those liposomes. The fourth and most often used method of incorporating proteins into liposomes involves the use of detergents. In such a method, the proteins and phospholipids are cosolubilized in a detergent to form micelles. The detergent is then removed, resulting in the spontaneous formation of bilayer vesicles with the protein incorporated therein. The detergent is incorporated into liposome as well as the protein and thus, these methods require removal of the detergent by methods such as dialysis, gel exclusion chromatography or adsorption on hydrophobic resins. The methods that use detergent are very slow because the detergent removal must be as complete as possible and also because a phase change that takes place during this process slows detergent removal even further. The detergent is also difficult to remove completely. Another disadvantage is that one cannot control the orientation of protein incorporated into the liposomes by using the detergent methods.

Current US Original Classification (1):

424/450

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#) [Previous Doc](#) [Next Doc](#) [Go to Doc#](#)



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Print

L3: Entry 10 of 28

File: USPT

Feb 1, 2000

US-PAT-NO: 6019998

DOCUMENT-IDENTIFIER: US 6019998 A

**** See image for Certificate of Correction ****

TITLE: Membrane structure

DATE-ISSUED: February 1, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
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US-CL-CURRENT: 424/450; 264/4.32, 264/4.33, 264/4.7, 427/213.33, 427/213.34,
427/214, 428/432, 428/433, 428/470

CLAIMS:

What is claimed is:

1. A stacked membrane structure having a spherical outer shape, comprising:

a first sub-structure;

a second sub-structure; and

a third sub-structure to an (N)th sub-structure;

wherein the (N)th sub-structure is an outermost sub-structure;

wherein N is an integer of at least three;

wherein each of said sub-structures comprises a layer having a bilayer membrane of lipid and a hydrophilic polymer part in aqueous phase;

wherein the lipid layer in a given substructure other than the (N)th sub-structure is between the hydrophilic polymer part of the given sub-structure and the hydrophilic polymer part of an adjacent sub-structure which is nearer to the (N)th sub-structure; and

wherein the hydrophilic polymer part in at least one sub-structure other than the (N)th sub-structure contains a first functional material, and the hydrophilic polymer part in at least another sub-structure other than said one sub-structure and other than the (N)th sub-structure contains a second

functional material, and the first functional material is different from the second functional material; and

wherein, of the sub-structures from said first sub-structure to the (N-1)th sub-structure, the composition of at least two sub-structures is different.

2. The structure according to claim 1, wherein the first or second functional compound is a functional organic compound.

3. The structure according to claim 2, wherein the functional organic compound is at least one material selected from the group consisting of enzyme, nucleic acid and pigment.

4. The structure according to claim 1, wherein the first or second functional compound is a functional inorganic compound.

5. The structure according to claim 4, wherein the functional inorganic compound is a pigment.

6. The structure according to claim 1, wherein the lipid layer in the sub-structure is in a fluid state in at least one of the sub-structures to maintain fluidity of the lipid membrane.

7. The structure according to claim 6, wherein the lipid membrane in the sub-structure in the fluid state is not cross-linked, or is partially cross-linked with the hydrophilic polymer part at specified functional groups at specified portions of the lipid membrane and the hydrophilic polymer part, in the same sub-structure.

8. The structure according to claim 1, wherein the hydrophilic polymer part in the sub-substructure is formed by radiant polymerization reaction of material for the hydrophilic polymer part.

9. The structure according to claim 1, wherein the hydrophilic part in each of the sub-structures contains a gel of water soluble polymers which are gelled by ion cross-linking.

10. The structure according to claim 9, wherein a hydrophilic polymer in the hydrophilic polymer part in the sub-structure does not exhibit a phase transition temperature close to a phase transition temperature for the lipid membrane to prevent breaking the lipid membrane.

11. The structure according to claim 1, further comprising an additional hydrophilic polymer layer surrounding a surface of the (N)th sub-structure.

12. A stacked membrane structure having a spherical outer shape, comprising:

a first sub-structure;

a second sub-structure; and

a third sub-structure to an (N)th sub-structure;

wherein the (N)th sub-structure is an outermost sub-structure;

wherein N is an integer of at least three;

wherein each of said sub-structures comprises a layer having a bilayer membrane of lipid and a hydrophilic polymer part in aqueous phase;

wherein the hydrophilic polymer part in a given sub-structure is between the lipid layer of the given sub-structure and the lipid layer of an adjacent sub-structure which is nearer to the (N)th sub-structure; and

wherein the lipid layer in at least one sub-structure other than the (N)th sub-structure contains a first functional material, and the lipid layer in at least another sub-structure other than said one sub-structure and other than the (N)th sub-structure contains a second functional material, and the first functional material is different from the second functional material; and

wherein, of the sub-structures from said first sub-structure to the (N-1)th sub-structure, the composition of at least two sub-structures is different.

13. The structure according to claim 12, wherein the first or second functional compound is a functional organic compound.

14. The structure according to claim 13, wherein the functional organic compound is at least one material selected from the group consisting of enzyme, nucleic acid and pigment.

15. The structure according to claim 12, wherein the first or second functional compound is a functional inorganic compound.

16. The structure according to claim 15, wherein the functional inorganic compound is a pigment.

17. The structure according to claim 12, wherein the lipid layer in the sub-structure is in a fluid state in at least one of the sub-structures to maintain fluidity of the lipid membrane.

18. The structure according to claim 17, wherein the lipid membrane in the sub-structure in the fluid state is not cross-linked, or is partially cross-linked with the hydrophilic polymer part at specified functional groups at specified portions of the lipid membrane and the hydrophilic polymer part in the same sub-structure.

19. The structure according to claim 12, wherein the hydrophilic polymer part in at least one of the sub-structures is formed by radiant polymerization reaction of material for the hydrophilic polymer part.

20. The structure according to claim 12, wherein the hydrophilic polymer part in at least one of the sub-structures contains a gel of water soluble polymers which are gelled by ion cross-linking.

21. The structure according to claim 20, wherein a hydrophilic polymer in the hydrophilic polymer part in the sub-structure does not exhibit a phase transition temperature close to a phase transition temperature for the lipid membrane to prevent breaking the lipid membrane.

22. The structure according to claim 12, further comprising an additional

hydrophilic polymer layer surrounding a surface of the (N)th sub-structure.

23. A stacked membrane structure comprising:

at least two structures, each of the structures comprising a lipid layer and a hydrophilic polymer part in aqueous phase,

wherein the hydrophilic polymer part in at least one of the structures disperses a particle which is a multilamellar liposome comprising,

a first sub-structure;

a second sub-structure; and

a third sub-structure to an (N)th sub-structure;

wherein the (N)th sub-structure is an outermost sub-structure;

wherein N is an integer of at least three;

wherein each of said sub-structures comprises a layer having a bilayer membrane of lipid and a hydrophilic polymer part in aqueous phase;

wherein the lipid layer in a given substructure other than the (N)th sub-structure is between the hydrophilic polymer part of the given sub-structure and the hydrophilic polymer part of an adjacent sub-structure which is nearer to the (N)th sub-structure; and

wherein the hydrophilic polymer part in at least one sub-structure other than the (N)th substructure contains a first functional material, and the hydrophilic polymer part in at least another sub-structure other than said one sub-structure and other than the (N)th sub-structure contains a second functional material, and the first functional material is different from the second functional material; and

wherein, of the sub-structures from said first sub-structure to the (N-1)th sub-structure, the composition of at least two sub-structures is different.

24. The structure according to claim 23, wherein the first or second functional compound is a functional organic compound.

25. The structure according to claim 24, wherein the functional organic compound is at least one material selected from the group consisting of enzyme, nucleic acid and pigment.

26. The structure according to claim 23, wherein the first or second functional compound is a functional inorganic compound.

27. The structure according to claim 26, wherein the functional inorganic compound is a pigment.

28. The structure according to claim 23, wherein the lipid layer in at least one of the sub-structures is in a fluid state to maintain fluidity of the lipid membrane.

29. The structure according to claim 28, wherein the lipid membrane in the sub-structure in the fluid state is not cross-linked, or is partially cross-linked with the hydrophilic polymer part at specified functional groups at specified portions of the lipid membrane and the hydrophilic polymer part in the same sub-structure.

30. The structure according to claim 23, wherein the hydrophilic polymer part in at least one of the sub-structure is formed by radiant polymerization reaction of material forming the hydrophilic polymer part.

31. The structure according to claim 23, wherein the hydrophilic polymer part in at least one of the sub-structure contains a gel of a water soluble polymer which is gelled by ion cross-linking.

32. The structure according to claim 31, wherein a hydrophilic polymer in the hydrophilic polymer part in the sub-structure does not exhibit a phase transition temperature close to a phase transition temperature for the lipid membrane in the sub-structure to prevent breaking the lipid membrane.

33. The structure according to claim 32, wherein the lipid membrane in the sub-structure in the fluid state is not cross-linked, or is partially cross-linked with the hydrophilic polymer part at specified functional groups at specified portions of the lipid membrane and the hydrophilic polymer part in the same sub-structure.

34. A stacked membrane structure comprising:

at least two structures, each of the structures comprising a lipid layer and a hydrophilic part in aqueous phase,

wherein the hydrophilic polymer part in at least one of the structures disperses a particle which is a multilamellar liposome comprising,

a first sub-structure;

a second sub-structure; and

a third sub-structure to an (N)th sub-structure;

wherein the (N)th sub-structure is an outermost sub-structure;

wherein N is an integer of at least three;

wherein each of said sub-structures comprises a layer having a bilayer membrane of lipid and a hydrophilic polymer part in aqueous phase;

wherein the hydrophilic polymer part in a given sub-structure other than the first sub-structure and the (N)th sub-structure is between the lipid layer of the given sub-structure and the lipid layer of an adjacent sub-structure which is nearer to the (N)th sub-structure; and

wherein the lipid layer in at least one sub-structure other than the (N)th sub-structure contains a first functional material, and the lipid layer in at least another sub-structure other than said one sub-structure and other than the (N)th sub-structure contains a second functional material, and the first

functional material is different from the second functional material; and

wherein, of the sub-structures from said first sub-structure to the (N-1)th sub-structure, the composition of at least two sub-structures is different.

35. The structure according to claim 34, wherein the first or second functional compound is a functional organic compound.

36. The structure according to claim 35, wherein the functional organic compound is at least one material selected from the group consisting of enzyme, nucleic acid and pigment.

37. The structure according to claim 34, wherein the first or second functional compound is a functional inorganic compound.

38. The structure according to claim 37, wherein the functional inorganic compound is a pigment.

39. The structure according to claim 34, wherein the lipid layer in at least one of the sub-structures is in a fluid state to maintain fluidity of the lipid membrane.

40. The structure according to claim 34, wherein the hydrophilic polymer part in at least one of the sub-structures is formed by radiant polymerization reaction of material for the hydrophilic polymer part.

41. The structure according to claim 34, wherein the hydrophilic polymer part in at least one of the sub-structures contains a gel of a water soluble polymer which is gelled by ion cross-linking.

42. The structure according to claim 41, wherein a hydrophilic polymer in the hydrophilic polymer part in the sub-structure does not exhibit a phase transition temperature close to a phase transition temperature for the lipid membrane in the sub-structure to prevent breaking the lipid membrane.

[Previous Doc](#)

[Next Doc](#)

[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

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L3: Entry 12 of 28

File: USPT

Sep 7, 1999

DOCUMENT-IDENTIFIER: US 5948756 A

TITLE: Therapeutic lipoprotein compositions

Brief Summary Text (110):

Alternately, the cholate-lipid dispersion method can be used to complex purified A and C apoproteins to egg PC liposomes containing cholesterol (Matz, 1982).

Current US Cross Reference Classification (2):424/450[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

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L3: Entry 17 of 28

File: USPT

Dec 29, 1998

DOCUMENT-IDENTIFIER: US 5853753 A

**** See image for Certificate of Correction ****

TITLE: Liposomes, method of preparing the same and use thereof in the preparation of drugs

Brief Summary Text (17):

Every bile acid derivative can be used according to the invention. Preferred bile acid derivatives are sodium cholate, sodium deoxycholate, sodium glycocholate, sodium taurocholate, sodium taurodeoxycholate, sodium ursocholate, and sodium chenoxocholate. The added bile acid and/or the derivatives thereof lead to a decrease in the surface tension of the liposomes and thus to an increased fusogenity which facilitates the fusion of the liposomes with body cells. It is also assumed that a presumably slight change in membrane potential which is more or less limited in time takes place on account of an interaction between surface tension and membrane potential after fusion of the liposomes with cells. For instance, Olson et al. (10) write that a weak depolarization of the transmembrane potential will take place a few minutes after rat liver cells have been treated with taurocholate. Furthermore, it might be that the membrane components, depending on their spatial arrangement, are subjected to certain spatial changes by the bile acid or the derivatives thereof, the changes being expressed in a changed permeability of the membrane or a changed production rate for "second messenger". The surfactant contained in the liposome must, however, not lead to a destabilization of either the liposome membrane, or, after fusion, of the cell membrane. Among all of the tested surfactants, bile acid and the derivatives thereof fulfill these requirements best. Moreover, it has been observed that the liposomes prepared in this way have been very stable for long periods of time.

Brief Summary Text (144):

As already described in sections I and II, the pain caused by viral infection or allergy disappears immediately in most cases after a liposome application. At least part of the mediators which are released by virally caused tissue lesions or allergic reactions are algogenic substances which alarm the nociceptors of the corresponding tissue. An objective assessment which furnishes proof of an action of physiological liposomes on nerve activity follows from the finding determined in Example 3 on the isolated intestine of a guinea pig. The pronounced inhibition of nerve activity in the case of relatively low liposome concentrations furnishes proof of the surprising efficacy of the cholate liposomes. The cholate-free liposomes used as a control lead to a stimulation of the nerves. Since an influence of the liposomes on the ion composition of the fluid is negligibly small due to the relatively small amounts of liposomes in the organ bath, but since cholate-free liposomes effect such a pronounced stimulation, it must be assumed that these liposomes, too, directly interact with the tissue or the cells. It is known from numerous experiments that surfactant-containing liposomes have a greater loss of enclosed hydrophilic small ions or molecules than surfactant-free liposomes. The cholate liposomes which are here used also have this property; i.e. surfactant-containing phospholipid membranes have a slightly increased permeability (in both directions) for such substances. When liposomes having a relatively great cholate amount (as the ones used here) fuse with cells, the entrained surfactants transmit the slightly increased membrane permeability, for instance for ions such as Na.sup.+ and K.sup.+, to the cell membranes. The membrane potential (resting

potential) is shifted on account of the now increased ion flow towards the "firing level" (10). Although this increases the excitability of the cell, the decrease in action potential corresponding to the depolarization simultaneously decreases the transmitter substances to be released (less vesicles are released), thereby effecting a reduced excitation of the postsynaptic cell (4). This presynaptic efficiency could be confirmed in further experiments with acetyl choline- or histamine-stimulated isolated guinea pig intestine. By contrast, a change in the membrane potential away from the "firing level" follows for a fusion of surfactant-free liposomes with cells, which consequently entails an increased action potential and an increased release (increased vesicle release) of transmitter substance. This leads to the observed stimulation of the intestinal contraction.

Brief Summary Text (165):

Since bile acids have an antibacterial effect, the cholate liposomes of the invention also exhibit bactericidal effects, at least against cholate-sensitive bacteria; i.e., some infections of bacterial etiology can also be treated in the case of the above-mentioned diseases.

Brief Summary Text (196):

1a. The incorporated cholate increases the permeability of the membrane because of the reduced surface tension above all for Na^+_{sup} (as the smallest one of the ions which are of importance to the membrane potential). This will lead to a slightly increased Na^+_{sup} inflow into the cell. Water follows the changed osmotic gradient and makes the cell swell until the inner pressure can withstand the influence. Moreover, cells within a tissue unit permit this swelling only to a very limited degree due to the cells adjacent thereto, i.e., only up to the size that is permitted by the intercellular space (30 nm on average). An increase in the cell diameter by about 1% can here roughly be expected. The swelling of the cells cannot be explained by the liposome volumes introduced into the cell after fusion, for in order to cope with this additional volume the cell would have to incorporate the more than hundredfold amount of liposomal membrane material, which cannot be imagined with a view to the cell physiology. This slight swelling means in vivo that the interstice decreases, which, in turn, makes it more difficult for substances (for instance, mediators) and particles (for instance viruses) to propagate. In accordance with the changed ion concentrations, the Na^+_{sup} -- K^+_{sup} -ATPase (Na^+ pump) will try under ATP consumption to re-establish the original conditions. Under normal conditions this pump process already needs about one third of the whole energy consumption of a cell (in electrically active nerve cells about two thirds (12)). Hence, the energy consumption and thus the total metabolism of a cell will presumably rise after interaction with physiological liposomes.

Brief Summary Text (197):

If the interaction of liposomes and cells corresponds to a fusion, the following events might take place: After fusion of the liposome membrane with the cell membrane, a membrane piece having a size of about $0.1 \mu\text{m}^2$ and increased permeability is incorporated through a liposome having a diameter of about 200 nm into the cell membrane ($500 \mu\text{m}^2$ outer membrane surface at a cell diameter of $12.6 \mu\text{m}$). As a result of this "new membrane piece" having a size of $0.1 \mu\text{m}^2$, there is an increase in the inflow and outflow of ions. However, since lipids, and presumably above all surfactants, diffuse laterally within the lipid double membrane at a rate of $2 \mu\text{m}/\text{sec}$ (7), the liposomal membrane piece already starts to flow apart at the time of the fusion, i.e., liposomal and cellular membrane components are mixed, which rapidly leads to a decrease in the ion permeability at this place of the cell membrane. A propagation of the liposomal membrane components to three to four times the area (0.3 to $0.4 \mu\text{m}^2$) might already lead to a substantial decline in the permeability (derived from experiments with liposomes having a lower cholate proportion). The high rate of lateral lipid diffusion of $2 \mu\text{m}/\text{sec}$ might therefore "seal" the originally $0.1 \mu\text{m}^2$ - sized liposomal membrane piece in fractions of a second for the greatest part. It

is not until an increased number of liposomes have fused with a cell that the surfactants introduced into the cell membrane lead to a change in permeability regarding the whole cell membrane. It can be shown in vitro that cells react negatively to a very short-time (15-30 min) offer of very high liposome concentrations. After a few hours, however, the cells will fully recover therefrom. This regeneration can, inter alia, be explained by the fact that 50% of the plasma membrane are internalized per hour (depending on the cell type) and that consequently half of the cell membrane is replaced every hour by membrane material from the interior of the cell. The proportion of the cell membrane in the total membrane material of a cell amounts to a few percent only (in a liver cell to about 2%). The original permeability of the cell membrane is therefore re-established within a relatively short period of time.

Detailed Description Text (4):

About 420 PFU (plaque forming units) of a fresh isolate of herpes simplex type 1 (1.2 ml) were incubated for different periods with a liposome suspension (1.2 ml) which contained 7.3.times.10.sup.13 liposomes per ml--unilamellar liposomes for the most part--after mixing. 3 g soybean lecithin were dissolved in 3 ml EtOH for preparing the liposomes. 3 g of sodium ascorbate, 0.27 g of common salt and 0.4 g of sodium cholate were dissolved in 23 ml double distilled water. The two solutions were well mixed by stirring, and the heterogenous mixture was sterilized by filtration at 5.times.10.sup.6 Pa. The resultant liposome dispersion was adjusted to pH 6.8 with 1N hydrochloric acid and diluted to the necessary test concentrations. The virus-liposome mixture was then added to a confluent monolayer of monkey kidney cells of the vero type for 15 minutes and subsequently replaced by medium. The evaluation of the PFU created by the lytic viruses was done after 72 hours. The results of the tests are shown in FIGS. 1a and 1b. The control was applied for time 0, i.e., viruses were added to the cells without liposomes. 420 PFU were measured after 3 days, which was equated with 100% PFU in the Fig. When the viruses were pipetted together with the liposomes and then immediately given to the cells, one only found 47% of the PFU in comparison with the control. This period of interaction of the liposomes with the viruses is about half a minute. After an incubation of the viruses with the liposomes for 10 minutes, only 26% PFU remained while less than 1% of the PFU could be detected after 30 minutes. About half of the viruses are directly inactivated after contact with the liposomes while some viruses remain infectious in the liposome suspension up to 30 minutes. This could be due to the heterogeneous morphology of the virus envelopes.

Detailed Description Text (46):

A piece of guinea pig ileum was electrically stimulated to contract in an organ bath, the intensity of the contractions were recorded and measured. Different concentrations of cholate-containing and surfactant-free liposomes (0.3 ml each) were added to the organ bath (25 ml buffer solution). The cholate-containing liposomes were prepared, as described in Experiment 1 of Example 1, while the surfactant-free liposomes were produced by treatment with ultrasonic waves of a lipid-containing, surfactant-free aqueous solution. As becomes apparent from FIG. 7, there is a huge difference in the effect of cholate-containing and surfactant-free liposomes. Cholate-containing liposomes (physiological liposomes) lead, in response to the concentration, to a decrease in the electrically stimulated intestinal contractions whereas the surfactant-free liposomes effect a considerable increase in the contraction.

Detailed Description Text (56):

5 g of pure soybean lecithin are dissolved in 5 ml ethanol. 0.9 g sodium cholate is dissolved in 85 ml of a 0.9% saline solution. Both solutions are filtered at room temperature at 5.times.10.sup.6 Pa through a filter with a pore size of 0.22 .mu.m. The resultant liposomal solution contains liposomes with a mean diameter of 130 nm. The liposome solution obtained after single filtration can optionally be adjusted to the desired pH value.

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424/450

Previous Doc

Next Doc

Go to Doc#

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

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Print

L3: Entry 18 of 28

File: USPT

Jan 7, 1997

DOCUMENT-IDENTIFIER: US 5591448 A

TITLE: Anti-viral therapeutic composition

Detailed Description Text (9):

The CD4 protein is incorporated into the bilayer of the liposome membrane. To start, unilamellar liposomes are formed by any known method. One way of obtaining the proper preparations is to pass a solution of detergent, phospholipids, and CD4 proteins through a gel chromatography column to remove the detergent. This method of liposome preparation yields unilamellar liposomes with most of the CD4 proteins oriented in a right-side-out direction. The method can only be used with detergents that have a high critical micelle concentration and a correspondingly low molecular weight of the micelles. Examples of these types of detergents are the bile sates sodium cholate, sodium deoxycholate, and the nonionic detergent beta-D-octylglucoside. These detergents can easily be removed from a phospholipid mixture by gel chromatography on SEPHADEX TM G 25 or G 50.

Current US Original Classification (1):424/450[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L3: Entry 27 of 28

File: USPT

Mar 15, 1988

DOCUMENT-IDENTIFIER: US 4731210 A

TITLE: Process for the preparation of liposomal medicaments

Detailed Description Text (60):

If necessary, the residual content of sodium cholate can be separated off by means of dialysis of the aqueous phase containing the liposomes or by gel chromatography.

Detailed Description Text (74):

Distearoyl phosphatidylcholine (DSPC) was converted into a lipid film with the addition of sodium cholate in a molar ratio of 0.2 as described, and the associates were formed at 60.degree. C. by adding 10 mM phosphate buffer of pH 7.1. The lipid concentration of the clear associate solution was 5 mg/ml. When this associate solution was allowed to cool slowly to room temperature, homogeneous unilamellar liposomes with a mean diameter of 260 nm were formed within 4 hours. When this jump in temperature was over a greater range, for example from 60.degree. C. to 4.degree. C., and was correspondingly more rapid, smaller homogeneous liposomes with a mean diameter of 75 nm resulted.

Detailed Description Text (76):

Phosphatidylcholine from egg yolk was converted into a lipid film with the addition of sodium cholate in a molar ratio of 0.72 as described, and the associates was formed at room temperature by adding 10 mM phosphate buffer of pH 7.1. The lipid concentration of the clear associate solution was 26 mg/ml. When the pH of this clear associate solution was reduced to pH 3 by rapidly adding 0.1N hydrochloric acid, the solubilizing agent was precipitated, homogeneous liposomes with a mean diameter of 30 nm being formed in the supernatant liquor.

Detailed Description Text (78):

100 mg of phosphatidylcholine from egg yolk together with 60 mg of sodium cholate, from which a molar ratio of lecithin to sodium cholate of 0.8 resulted, was converted into a lipid film as described, and the associates were formed at room temperature by adding 5 ml of 10 mM phosphate buffer of pH 7.1. 49.75 mg of phosphatidylcholine were lyophilized in a second vessel. The associate solution (5 ml) described above was added instantaneously to this highly disperse lyophilisate, with stirring; liposomes with a mean diameter of 70 nm spontaneously formed as a result of the change in the molar ratio of bilayer-forming substance to solubilizing agent from originally 0.8 to 1.2.

Detailed Description Paragraph Table (1):

TABLE

SOLUBILIZING BILAYER-FORMING SUBSTANCE AGENT MOLAR RATIO MODEL SUBSTANCE

Concentration in the aqueous Starting concentration Bilayer-forming agent/ Amount employed, based on phase during associate formation (mg/ml) (mg/ml) solubilizing agent the aqueous phase

(mg/ml)

EYL/DSPC Na <u>cholate</u> 0.8 -- 3:1.sup.(* .sup.)	3.34	9.11	9.75	EYL/DSPC C.sub.8 - glycoside 0.2 -- 3:1.sup.(* .sup.)	3.34	24.75	9.75	DSPC Na <u>cholate</u> 0.2 -- 15.81
43.05 EYL C.sub.7 -glycoside 0.13	cholesterol 8.0	26,67	0.81	EYL/choleste- C.sub.8 -glycoside 0.25	6-carboxy- rol.sup.(1)	fluorescein 5:1.sup.(* .sup.)	1.73	50.0

37.63 10.4

LIPOSOME

SIZE INCORPORATION RATE EXPERIMENTAL DILUTION RATIO Diameter (nm) Model substance
in % of the starting CONDITIONS

1:5 60.5

-- Associate solution pre- pared at 56.degree. C. Dilution at room temperature or
56.degree. C. 1:5 126 -- Associate solution pre- pared at 56.degree. C. Dilution at
room temperature or 56.degree. C. 1:5 91.4 -- Associate solution pre- pared at
60.degree. C. Dilution at 60.degree. C. 1:3 99 80 Associate solution pre- pared at
room temperature. Dilution at room temperature.sup.(2) 1:4 190 3 Associate solution
pre- pared at 55.degree. C. Dilution at 55.degree. C..sup.(3)

.sup.(1)

In this case, cholesterol is to be regarded as a component of th membrane .sup.(2)

Non-bonded model substance separated off by means of gel chromatography or

dialysis .sup.(3) Non-bonded model substance separated off by means of gel

chromatography .sup.(*.sup.) Molar ratio Abbreviations EYL = egg lecithin DSPC =

distearoyl phosphatidylcholine C.sub.8 -glycoside = 10-n-octyl .beta.-

Dglucopyranoside C.sub.7 -glycoside = 10-n-heptyl .beta.-Dglucopyranoside

Current US Cross Reference Classification (3):

424/450

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#) [Previous Doc](#) [Next Doc](#) [Go to Doc#](#)☐ [Generate Collection](#) [Print](#)

L3: Entry 7 of 28

File: USPT

Apr 16, 2002

DOCUMENT-IDENTIFIER: US 6372720 B1

TITLE: Liposome fusion and delivery vehicle

Detailed Description Text (55):

In an embodiment which includes a ligand for cell targeting, the following procedures can be used. First, a cross-linking phosphatidylethanolamine with a disulfide linkage is made. DOPE is reacted with a five-fold excess of the homobifunctional cross-linking reagent DTSP (3,3'-dithio-bis(propionic acid N-hydroxysuccinimide ester)) in chloroform in the presence of triethylamine. The resulting conjugate is then purified by LH-20 Sephadex chromatography. This conjugate will react with free amino groups of targeting ligands when carried out in a detergent solution (100 mM cholate, 10 mM borate buffer, pH 9.2) for several hours at room temperature. This solution is then mixed with the remainder of the liposome components prior to dialysis.

Detailed Description Text (66):

This procedure is used when the nuclear localization signal peptide (or other positively charged composition) plus the N-iodoacetyl DOPE (or other cross-linking lipid) are used immediately for liposome formation. In a separate embodiment, after the 3-4 hours reaction in dimethylformamide, the lipid-peptide conjugate is precipitated with 5 ml of tert-butyl methyl ether. The precipitate is centrifuged a 1000.times.g, and the supernatant with the excess (unreacted) lipid is discarded. The precipitate, containing lipid-peptide conjugate without excess lipid, is redissolved in dimethylsulfoxide and stored at -20 degrees C. until further use. When used for liposome formation, the lipid-peptide conjugate in dimethylsulfoxide is added directly to the solution of 250 mM cholate in PB buffer.

Detailed Description Text (135):

A conjugate of DOPE and polyethylene glycol polymer (average molecular weight 5000) containing a disulfide linkage was prepared according to methods described in Kirpotin, et al., FEBS Letters, 388(2-3):115-8 (1996). The conjugate was stored at -20 degrees in ethanol at a concentration of approximately 2 micromoles per milliliter. Liposomes were then prepared as described in specific example 2 with the exception that a 3-fold greater volume of 250 mM cholate in PB buffer was used in each step. The liposomes were prepared with the following compositions: 135 nmol of nuclear localization signal peptide (SEQ ID NO: 6) with each molecule of nuclear localization signal peptide coupled to two molecules of N-iodoacetyl DOPE via the two cysteine amino acids; 2100 nmol of 14:1 PC; and 210 nmol fusogenic peptide (SEQ ID NO:3). To one preparation was added 70 nmol of DOPE-polyethylene glycol conjugate, at the same point where the 14:1 PC was added to the preparation. A separate preparation was made without polymer.

Current US Cross Reference Classification (1):

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